Remarks

Claims 1-16 were pending in the subject application. By this Amendment, claims 7-12 and 16 have been amended, claims 1-6 and 13-15 have been cancelled, and new claims 17-34 have been added. The undersigned avers that no new matter is introduced by this amendment. Entry and consideration of the amendments presented herein is respectfully requested. Accordingly, claims 7-12 and 16-34 are currently before the Examiner for consideration. Favorable consideration of the pending claims is respectfully requested.

As an initial matter, the applicant notes that the supplemental Information Disclosure Statement (IDS) submitted on May 27, 2004 was not acknowledged in the instant Office Action. The applicant reviewed the status of the subject application on the U.S. Patent Office's Patent Application Information Retrieval (PAIR) system and found that the Patent Office has received the supplemental IDS. The applicant respectfully requests that the Examiner consider the reference listed on the Form PTO/SB/08 and make its consideration of record in the subject application.

The applicant notes that the box next to number 10 on the Office Action Summary sheet is checked, but the box indicating whether the drawings are accepted or objected to by the Examiner is not checked. Upon inquiring with the Examiner telephonically on November 4, 2004, the undersigned was informed that the drawings are accepted.

The Examiner indicates that the filing date listed by the applicant (February 13, 2002) does not match the filing date listed in the Patent Office database (February 15, 2002). The applicant respectfully submits that February 13, 2002 is the date the subject national application was deposited with the Patent Office via Express Mail and is not necessarily the date in which all requirements under 35 U.S.C. §371 were satisfied.

By this Amendment, claims 7-12 and 16 have been amended to delete reference to RhoB variants. Claim 16 has been amended to recite that an effective amount of a RhoB protein, or a pharmaceutically acceptable salt thereof, is administered. Support for this amendment can be found, for example, at page 7, lines 26-31, and page 8, lines 1-5, of the specification, and the claims as originally filed. Support for claims 17 and 27 can be found, for example, at page 3, lines 26-31, page 4, lines 1-11, page 5, lines 23-25, page 7, lines 1-5 and 26, page 17, lines 25-28, of the specification,

and the claims as originally filed. Support for claim 18 can be found, for example, at page 7, lines 18-19, and page 8, lines 10-11, of the subject specification. Support for claims 19 and 20 can be found, for example, at page 3, lines 26-31, page 4, lines 1-11, page 5, lines 23-25, page 7, lines 1-5 and 26, page 17, lines 25-28, of the specification, and the claims as originally filed. Support for claim 21 can be found, for example, at page 8, lines 6-11, page 10, lines 6-9, page 11, lines 5-18, and page 16, lines 24-32, of the specification. Support for claims 22 and 29 can be found, for example, at page 8, lines 12-14, of the specification. Support for claims 23 and 34 can be found, for example, at page 6, lines 5-6, of the subject specification. Support for claims 24 and 31 can be found, for example, at page 6, lines 28-32, of the specification, and the claims as originally filed. Support for claims 25 and 32 can be found, for example, at page 5, line 32, of the subject specification. Support for claims 26 and 33 can be found, for example, at page 5, lines 26-32, and page 6, lines 1-6, of the specification.

Claim 5 has been rejected under 35 U.S.C. §112, second paragraph, as indefinite. By this Amendment, the applicant has cancelled claim 5, rendering this rejection moot. New claims 25 and 32 recite that the cancerous cells are cells of a solid tumor. The applicant respectfully submits that the metes and bounds of the claimed subject matter is clear to those of ordinary skill in the art. Reconsideration and withdrawal of the rejection under 35 U.S.C. §112, second paragraph, is respectfully requested.

Claims 1-5, 8-12, and 16 have been rejected under 35 U.S.C. §112, first paragraph, as lacking sufficient written description. The Office Action indicates that the subject application does not indicate the distinguishing attributes identifying members of the genus comprising RhoB variants. The applicant respectfully submits that the subject specification provides a sufficient written description of the subject matter of claims 1-5, 8-12, and 16. However, by this Amendment, the applicant has cancelled claims 1-5 and amended claims 7-12 and 16 to remove the term "RhoB variants". The term "RhoB variants" is not recited in new claims 17-33. Accordingly, reconsideration and withdrawal of the rejection under 35 U.S.C. §112, first paragraph, is respectfully requested.

Claims 1-5, 7-12, and 16 have been rejected under 35 U.S.C. §112, first paragraph, as non-enabled by the subject specification. The applicant respectfully traverses and submits that the claims are fully enabled by the subject specification.

The Office Action indicates that the subject specification does not provide sufficient guidance resolving issues associated with *in vivo* delivery of RhoB protein and nucleotides encoding RhoB, and treatment effects. Submitted herewith for the Examiner's consideration is Exhibit B, which provides further evidence for the potent tumor suppressive activity of RhoB. The experimental data presented in Exhibit B demonstrates that a nucleic acid construct encoding RhoB can be delivered within a human lung tumor growing in a mouse model, resulting in inhibited tumor growth. Previously, Dr. Sebti's laboratory showed that forced expression of RhoB in human cancer cells suppresses tumor growth *ex vivo*. As described at pages 16 and 17 of the subject application, this was done by stably expressing RhoB in human pancreatic cancer cells (Panc-1 cells), subcutaneously injecting these cells into mice, and showing that the RhoB-expressing human cancer cells did not grow. This previous experiment provided evidence for the potent tumor suppressive activity of RhoB. The next objective was to demonstrate that tumor suppression can be achieved by delivering RhoB *in vivo* directly into growing tumors. To this end, under the direction of Dr. Sebti, wild-type RhoB was cloned into a viral vector, as taught at page 7 of the subject application, and subcutaneously injected human lung cancer A-549 cells into the flanks of female athymic nude mice, using the mice as a tumor xenograft model. When tumors reached an average volume of 150-200 mm^3, approximately $5x10^{10}$ adenoviral particles expressing adenoviral-RhoB, adeno-RhoA, or adenovector (vector alone), were injected everyday intra-tumorally for 12 days (150 µl per injection). As is evident from the graph (Exhibit B), A-549 tumors injected intra-tumorally with vector alone grew from 200 mm^3 to 800 mm^3 over a period of 17 days. In contrast, A-549 tumors injected with adeno-RhoB grew to only 400 mm^3. Tumors injected with adeno-RhoA grew to 900 mm^3. Thus, in this experiment, it was confirmed that injection of human tumors in nude mice *in vivo* with adeno-RhoB results in reduced tumor growth. Furthermore, based on the *in vitro* and *in vivo* experimental data provided at pages 13-19 of the subject application, which characterize the mechanism of RhoB's potent tumor suppressor function, it is reasonable to expect that the following underlying physiological effects occur *in vivo*, commensurate with suppressed or inhibited tumor growth:

induced apoptosis of transformed cells; inhibited oncogenic signaling of cells; and decreased phosphorylation of Akt, Erk1, and Erk2 proteins within transformed cells. The applicant respectfully submits that one of ordinary skill in the art would accept the *in vitro* and *in vivo* data presented in the subject application and Exhibit B as reasonably predictive of RhoB's therapeutic benefit (*e.g.*, suppression of cancer cell growth) in mammals, including humans.

The nude mouse tumor xenograft model is a well recognized animal model of cancer. The nude mouse tumor xenograft model utilized in the above-described experiment represents a stringent model for assessment of the therapeutic potential of RhoB. Human tissue xenograft models are currently recognized by those in the field as one of the best tools for conducting pre-clinical *in vivo* analyses of intact human tissue. The nude mouse tumor xenograft model reflects the clinical situation in that the great majority of anti-cancer agents found to be clinically active in human subjects was similarly active in the tumor xenograft mouse model. Therefore, one of ordinary skill in the art would expect that the experimental results obtained using this model would reasonably correlate with a therapeutic benefit in human patients.

In addition to adenovector-mediated gene delivery, other techniques for RhoB delivery may be employed, as taught in the subject application. For example, other viral and non-viral vectors may be used to deliver nucleic acid constructs encoding RhoB to cancerous cells, resulting in RhoB expression. Furthermore, tissue-specific promoters may be used, or as taught at page 7, lines 18-25, of the specification, event-specific promoters may be utilized with nucleic acid constructs encoding RhoB to further optimize and localize expression within the diseased tissues. Submitted herewith for the Examiner's consideration is the Robson *et al.* publication, which reviews various methodologies and vectors available for delivering and expressing a polynucleotide *in vivo* for the purpose of treating cancer (Robson, T. Hirst, D.G., *J. Biomed. and Biotechnol.*, 2003, 2003(2):110-137). Among the various targeting techniques available, transcriptional targeting using tissue-specific and event-specific transcriptional control elements is discussed. For example, Table 1 at page 112 of the Robson *et al.* publication lists several tissue-specific promoters useful in cancer therapy, many of which were available at the time the patent application was filed. Tables 2-4 of the Robson *et al.* publication list tumor-specific promoters, tumor environment-specific promoters, and exogenously controlled inducible promoters, many of which were available at the time the patent

application was filed. The successful delivery and expression of the p53 tumor suppressor gene *in vivo* has been documented (Horowitz, J. *Curr. Opin. Mol. Ther.*, 1999, 1(4):500-509; Von Gruenigen, V.E. *et al. Int. J. Gynecol. Cancer*, 1999, 9(5):365-372; Fujiwara, T. *et al., Mol. Urol.*, 2000, 4(2):51-54, respectively). In view of the means available for delivery and expression of a polynucleotide to a human or non-human mammal *in vivo*, and the success demonstrated with these systems, one of ordinary skill in the art would expect that the obstacles to RhoB gene delivery set forth in the Office Action can be addressed by optimization, rather than undue experimentation.

The Office Action indicates that there are many unresolved problems in the art of protein delivery, such as enzymatic degradation of the protein, cell uptake, toxicity to normal cells, *etc.* With respect to toxicity to normal cells, it should be noted that there is no experimental data within the subject application that suggests an abundance of RhoB would be toxic to normal cells. Thus, like the tumor suppressor p53, non-specific delivery of the RhoB gene or protein should not compromise its clinical benefits. The applicant acknowledges that while the issues raised in the Office Action are considerations relevant in the selection of any protein delivery method, reagents and methods for delivering small molecules and proteins to the interior of cells do exist, and most if not all of these issues can be avoided or minimized. For example, peptides known as "cell penetrating peptides" (CPP) or "protein transduction domains" (PTD) have an ability to cross the cell membrane and enter the cell. PTDs can be linked to a cargo moiety such as a drug, peptide, or full-length protein, and can transport the moiety across the cell membrane. One well characterized PTD is the human immunodeficient virus (HIV)-1 Tat peptide (see, for example, Frankel *et al.*, U.S. Patent Nos. 5,804,604; 5,747,641; 6,674,980; 5,670,617; and 5,652,122; Fawell, S. *et al., Proc. Natl. Acad. Sci.* U.S.A., 1994, 91:664-668). Peptides such as the homeodomain of *Drosophila* antennapedia (ANTp) and arginine-rich peptides display similar properties (Derossi, D. *et al., J. Biol. Chem.*, 1994, 269:10444-10450; Derossi, D. *et al., Trends Cell Biol.*, 1998, 8:84-87; Rojas, M. *et al., Nat. Biotechnol.*, 1998, 16:370-375; Futaki, S. *et al., J. Biol. Chem.*, 2001, 276:5836-5840). VP22, a tegument protein from Herpes simplex virus type 1 (HSV-1), also has the ability to transport proteins across a cell membrane (Elliot *et al., Cell*, 1997, 88:223-233; Schwarze S.R. *et al., Trends Pharmacol. Sci.*, 2000, 21:45-48). A common feature of these carriers is that they are highly basic and hydrophilic (Schwarze S.R. *et al., Trends Cell Biol.*, 2000, 10:290-295). Coupling of these

carriers to marker proteins such as beta-galactosidase has been shown to confer efficient internalization of the marker protein into cells. More recently, chimeric, in-frame fusion proteins containing these carriers have been used to deliver proteins to a wide spectrum of cell types both *in vitro* and *in vivo*. For example, VP22-p53 chimeric protein retained its ability to spread between cells and its pro-apoptotic activity, and had a widespread cytotoxic effect in p53 negative human osteosarcoma cells *in vitro* (Phelan, A. *et al.*, *Nature Biotechnol.*, 1998, 16:440-443). Intraperitoneal injection of the beta-galactosidase protein fused to the HIV-1 Tat peptide resulted in delivery of the biologically active fusion protein to all tissues in mice, including the brain (Schwarze S.R. *et al.*, *Science*, 1999, 285:1569-1572). Liposomes of various compositions can also be used for site-specific delivery of proteins and drugs (Witschi, C. *et al.*, *Pharm. Res.*, 1999, 16:382-390; Yeh, M.K. *et al.*, *Pharm. Res.*, 1996, 1693-1698). The interaction between the liposomes and the protein cargo usually relies on hydrophobic interactions or charge attractions, particularly in the case of cationic lipid delivery systems (Zelphati, O. *et al.*, *J. Biol. Chem.*, 2001, 276:35103-35110). Tat peptide-bearing liposomes have also been constructed and used to deliver cargo directly into the cytoplasm, bypassing the endocytotic pathway (Torchilin V.P. *et al.*, *Biochim. Biophys. Acta-Biomembranes*, 2001, 1511:397-411; Torchilin V.P. *et al.*, *Proc. Natl. Acad. Sci.* USA, 2001, 98:8786-8791). When encapsulated in sugar-grafted liposomes, pentamidine isethionate and a derivative have been found to be more potent in comparison to normal liposome-encapsulated drug or to the free drug (Banerjee, G. *et al.*, *J. Antimicrob. Chemother.*, 1996, 38(1):145-150). A thermo-sensitive liposomal taxol formulation (heat-mediated targeted drug delivery) has been administered *in vivo* to tumor-bearing mice in combination with local hyperthermia, and a significant reduction in tumor volume and an increase in survival time was observed compared to the equivalent dose of free taxol with or without hyperthermia (Sharma, D. *et al.*, *Melanoma Res.*, 1998, 8(3):240-244). Topical application of liposome preparations for delivery of insulin, IFN-alpha, IFN-gamma, and prostaglandin E1 have met with some success (Cevc G. *et al.*, *Biochim. Biophys, Acta*, 1998, 1368:201-215; Foldvari M. *et al.*, *J. Liposome Res.*, 1997, 7:115-126; Short S.M. *et al.*, *Pharm. Res.*, 1996, 13:1020-1027; Foldvari M. *et al.*, *Urology*, 1998, 52(5):838-843; U.S. Patent No. 5,853,755). Antibodies represent another targeting device that may make liposome uptake tissue or cell-specific (Mastrobattista, E. *et al.*, *Biochim. Biophys. Acta*, 1999, 1419(2):353-363; Mastrobattista, E. *et al.*, *Adv. Drug Deliv. Rev.*,

1999, 40(1-2):103-127). The liposome approach offers several advantages, including the ability to slowly release encapsulated drugs and proteins, the capability of evading the immune system and proteolytic enzymes, and the ability to be targeted to tumors and preferentially accumulate in tumor tissues and their metastases by extravasation through their leaky neovasculature. Other carriers have also been used to deliver anti-cancer drugs to neoplastic cells, however, such as polyvinylpyrrolidone nanoparticles and maleylated bovine serum albumin (Sharma, D. *et al.*, *Oncol. Res.*, 1996, 8(7-8):281-286; Mukhopadhyay, A. *et al.*, *FEBS Lett.*, 1995, 376(1-2):95-98). Thus, using targeting and encapsulation technologies, which are very versatile and amenable to rational design and modification, and which were available at the time the patent application was filed, the obstacles to RhoB protein delivery cited in the Office Action can be eliminated or minimized to a large extent. Furthermore, because many liposome compositions are also viable delivery vehicles for genetic material, many of the advantages of liposomes are equally applicable to nucleic acid sequences encoding RhoB.

The Office Action indicates that the subject application lacks guidance for the method of combining RhoB therapy with an additional therapy, such as chemotherapy or radiotherapy. Combination therapies, such as a combination of chemotherapy, radiotherapy, and/or surgery, have been utilized for the treatment of cancer for some time. Likewise, the applicant respectfully submits that there would be no technological hurdle in using one or more of these techniques in combination with RhoB therapy of the invention. Commonly, several chemotherapy drugs are combined (combination chemotherapy). The rationale for combination chemotherapy is to use drugs that work on different parts of the cancer cell's life cycle, thereby increasing the likelihood more cancer cells will be killed. For some cancers, the best approach is a combination or sub-combination of surgery, radiation, and chemotherapy. Surgery or radiation therapy treats cancer that is confined locally, while chemotherapy also kills cancer cells that may have spread. Sometimes, radiation or chemotherapy is given before surgery to reduce the size of a tumor, thereby making the complete removal of the tumor using surgery more likely, or after surgery to destroy any remaining cancer cells. The stage of the cancer also often determines whether single therapy or a combination is indicated. For example, locally advanced breast cancer is usually treated with chemotherapy, radiation therapy, and surgery. Sometimes, combination chemotherapy is used not to cure but to

reduce symptoms and prolong life. Combination chemotherapy can be useful for people with advanced cancers that are not suitable for irradiation or surgical treatment. As the Examiner is aware, the specification need not disclose what is well-known to those skilled in the art and preferably omits that which is well-known to those skilled and already available to the public. *In re Buchner*, 18 USPQ2d 1331, 1332 (Fed. Cir. 1991); *Hybritech Inc. v. Monoclonal Antibodies, Inc.*, 231 USPQ 81, 94 (Fed. Cir. 1986), *cert. denied*, 480 U.S. 947 (1987); and *Lindemann Maschinenfabrik GMBH v. American Hoist & Derrick Co.*, 221 USPQ 481, 489 (Fed. Cir. 1984).

At pages 8 and 9 of the Office Action, the Examiner raises several issues such as toxicity to normal tissues upon administration, route of delivery, the effect of the immune response, doses to be administered, dose schedules, *etc.* The applicant respectfully submits that an application for patent is not required to show that a claimed method of treatment of a disease condition results in a cure of that disease condition, or even that clinical efficacy is achieved. The Federal Circuit has made it clear that the showing for therapeutic utility that is sufficient to satisfy the patent laws is not to be confused or equated with the showing required by the Food & Drug Administration for drugs, medical devices, and procedures. *Scott v. Finney*, 32 USPQ2d 1115 (Fed. Cir. 1994) and Manual of Patent Examining Procedure 2164.05. Given the state of the art as demonstrated by the scientific publications submitted herewith, and the information provided in the subject specification and the experimental results obtained therewith, one of ordinary skill in the art can deliver RhoB to cancerous cells *in vitro* or *in vivo*, without resort to undue experimentation. Thus, the applicant respectfully submits that the subject specification enables the methods and compositions as currently claimed.

At page 10 of the Office Action, the Examiner raises several issues regarding preventing malignant transformation, which is recited in claim 11 of the subject application. The applicant respectfully submits that the claimed subject matter is fully enabled by the specification. Because it has been shown that cancer-causing genes such as Ras, EGFR, and ErbB2 down-regulate RhoB expression to promote tumorigenesis and demonstrate that ectopic expression of RhoB blocks Ras, EGFR, and ErbB2-mediated malignant transformation, the applicant submits that one of ordinary skill in the art would reasonably expect that introduction of RhoB into normal cells can prevent malignant transformation.

The applicant respectfully submits that, in view of the disclosure of the subject specification as originally filed, and in view of the experimental results developed using those techniques which are described in the specification and known to those of ordinary skill in the art, methods for delivering RhoB to cancerous cells *in vitro* or *in vivo* are fully enabled.

Accordingly, the applicant respectfully submits that, given the teaching of the specification and the state of the art, one of ordinary skill in the art could carry out the claimed methods without the need for undue experimentation. In view of the foregoing remarks, reconsideration and withdrawal of the rejection under 35 U.S.C. §112, first paragraph, is respectfully requested.

Claims 1-2, 5, and 7-11 has been rejected under 35 U.S.C. §102(a) as being anticipated by Chen *et al.* (*J. Biol. Chem.*, 2000, 275:17974-17978). The applicant respectfully traverses this ground for rejection and submits that the Chen *et al.* publication is not prior art to the claimed invention.

As the Examiner is undoubtedly aware, the requirements for authorship and inventorship are not the same. The inventorship of the claimed invention and the authorship of the Chen *et al.* publication differ in that although Zhi Chen, Jiazhi Sun, Anne Pradines, Gilles Favres, and Jalila Adnane are co-authors of the Chen *et al.* publication, they are not named as inventors on the subject application.

Submitted herewith is a Declaration under 37 C.F.R. § 1.132 by Dr. Said M. Sebti for the Examiner's consideration. Dr. Sebti explains in his Declaration that although Zhi Chen, Jiazhi Sun, Anne Pradines, Gilles Favres, and Jalila Adnane are acknowledged as co-authors on the Chen *et al.* publication, they did not contribute to the conception of the invention claimed in the subject application. Therefore, despite their helpful research assistance, they were not included as co-inventors on the subject application.

The subject matter pertaining to the claimed invention that is described within the Chen *et al.* publication was invented by the named inventor, *i.e.*, Dr. Said M. Sebti. Therefore, the Chen *et al.* publication represents the inventor's own disclosure of his invention published less than one year prior to the filing date of the subject application.

"[O]ne's own invention, whatever the form of disclosure to the public, may not be prior art against oneself, absent a statutory bar." *In re Facius,* 161 USPQ 294, 301 (CCPA 1969); and MPEP

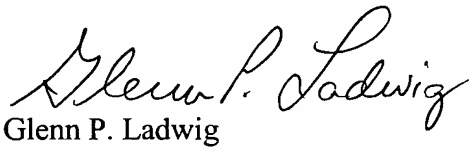
§715.01(c). Therefore, under the authority of *In re Facius*, the disclosure contained in the Chen *et al.* publication cannot be used as a reference against the applicant's claimed invention. Accordingly, reconsideration and withdrawal of the rejection under 35 U.S.C. §102(a) is respectfully requested.

In view of the foregoing remarks and amendments to the claims, the applicant believes that the currently pending claims are in condition for allowance, and such action is respectfully requested.

The Commissioner is hereby authorized to charge any fees under 37 C.F.R. §§ 1.16 or 1.17 as required by this paper to Deposit Account 19-0065.

The applicant invites the Examiner to call the undersigned if clarification is needed on any of this response, or if the Examiner believes a telephonic interview would expedite the prosecution of the subject application to completion.

Respectfully submitted,



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GPL/mv
Attachments: Petition and Fee for Extension of Time
Amendment Transmittal Letter
Declaration by Dr. Said Sebti under 37 C.F.R. §1.132, including Exhibits A-B
Robson *et al.* publication (*J. Biomed. and Biotechnol.*, 2003, 2003(2):110-137)